

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : James S. Norris et al. Art Unit : 1633
Serial No. : 10/082,973 Examiner : Janet L. Epps-Ford
Filed : February 26, 2002 Conf. No. : 8113
Title : TISSUE-SPECIFIC AND TARGET RNA-SPECIFIC RIBOZYMES

Mail Stop Amendment

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF JAMES S. NORRIS UNDER 37 C.F.R. § 1.131

I, James S. Norris, hereby declare as follows:

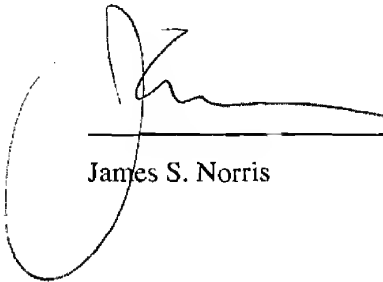
1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.
2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).
3. The Norris *et al.* reference lists its publication date as June 11, 1998.
4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, Gary A. Clawson, Michael G. Schmidt, Brian D. Hoel, Wei-Hua Pan, Joseph W. Dolan, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under Gary A. Clawson's supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP

sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

5. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

6/11/05
Date

60427105.doc


James S. Norris

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant :	James S. Norris et al.	Art Unit :	1633
Serial No. :	10/082,973	Examiner :	Janet L. Epps-Ford
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P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF GARY A. CLAWSON UNDER 37 C.F.R. § 1.131

I, Gary A. Clawson, hereby declare as follows:

1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.
2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).
3. The Norris *et al.* reference lists its publication date as June 11, 1998.
4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, James S. Norris, Michael G. Schmidt, Brian D. Hoel, Wei-Hua Pan, Joseph W. Dolan, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under my supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

Applicant : James S. Norris et al.
Serial No. : 10/082,973
Filed : February 26, 2002
Page : 2 of 2

Attorney's Docket No.: 14017-004002 / PSU 96-
1566

5. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

JUNE 11, 2007
Date

Gary A. Clawson
Gary A. Clawson

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	: James S. Norris et al.	Art Unit	: 1633
Serial No.	: 10/082,973	Examiner	: Janet L. Epps-Ford
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Mail Stop Amendment

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P.O. Box 1450

Alexandria, VA 22313-1450

DECLARATION OF MICHAEL G. SCHMIDT UNDER 37 C.F.R. § 1.131

I, Michael G. Schmidt, hereby declare as follows:

1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.

2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).

3. The Norris *et al.* reference lists its publication date as June 11, 1998.

4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, James S. Norris, Gary A. Clawson, Brian D. Hoel, Wei-Hua Pan, Joseph W. Dolan, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under Gary A. Clawson's supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

5. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.



11 June 2007

Date

Michael G. Schmidt

Dr Schmidt's signature page.doc

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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Alexandria, VA 22313-1450

DECLARATION OF BRIAN D. HOEL UNDER 37 C.F.R. § 1.131

I, Brian D. Hoel, hereby declare as follows:

1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.

2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).

3. The Norris *et al.* reference lists its publication date as June 11, 1998.

4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, James S. Norris, Michael G. Schmidt, Gary A. Clawson, Wei-Hua Pan, Joseph W. Dolan, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under Gary A. Clawson's supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

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June 10 '07

Date

Brian D. Hoel

Brian D. Hoel

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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Alexandria, VA 22313-1450

DECLARATION OF WEI-HUA PAN UNDER 37 C.F.R. § 1.131

I, Wei-Hua Pan, hereby declare as follows:

1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.

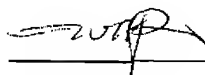
2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).

3. The Norris *et al.* reference lists its publication date as June 11, 1998.

4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, James S. Norris, Michael G. Schmidt, Brian D. Hoel, Gary A. Clawson, Joseph W. Dolan, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under Gary A. Clawson's supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

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6-11-07
Date


Wei-Hua Pan

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06/11/2007 14:39 7924882

MICRO

PAGE 02/03

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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Commissioner for Patents
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Alexandria, VA 22313-1450

DECLARATION OF JOSEPH W. DOLAN UNDER 37 C.F.R. § 1.131

I, Joseph W. Dolan, hereby declare as follows:

1. I am an inventor of currently pending claims 39-55 of the above-referenced patent application.

2. In an Office Action dated January 11, 2007, claims 39, 43-48, 50-51, and 53-55 were rejected under 35 U.S.C. § 102(a) as allegedly being anticipated by Norris *et al.* (WO98/24925).

3. The Norris *et al.* reference lists its publication date as June 11, 1998.

4. Prior to June 11, 1998, and thus necessarily before the publication date of the Norris *et al.* reference, James S. Norris, Michael G. Schmidt, Brian D. Hoel, Wei-Hua Pan, Gary A. Clawson, and I worked together in this country to complete the conception of the invention recited in present claims 39, 43-48, 50-51, and 53-55 of the above-referenced patent application, and to reduce said invention to practice, as evidenced by a copy of pages from Ping Xin's laboratory notebook and pages from Shani Schalles' laboratory notebook, each produced under Gary A. Clawson's supervision, which is attached as Exhibit A. The pages include multiple examples of pCHOP sequences. The dates on these pages, all of which are prior to June 11, 1998, have been blacked out.

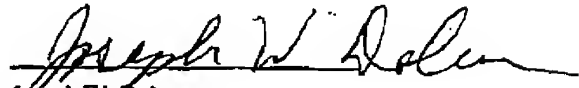
Jun 13 2007 8:16PM HP LASERJET FAX

p.3

5. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

6/13/07

Date


Joseph W. Dolan

60427107.doc



Signal G:354 A:393 T:181 C:274
DT4%Ac{A Set-AnyPrimer}

Page 1 of 3
9:42 AM

Data Analysis

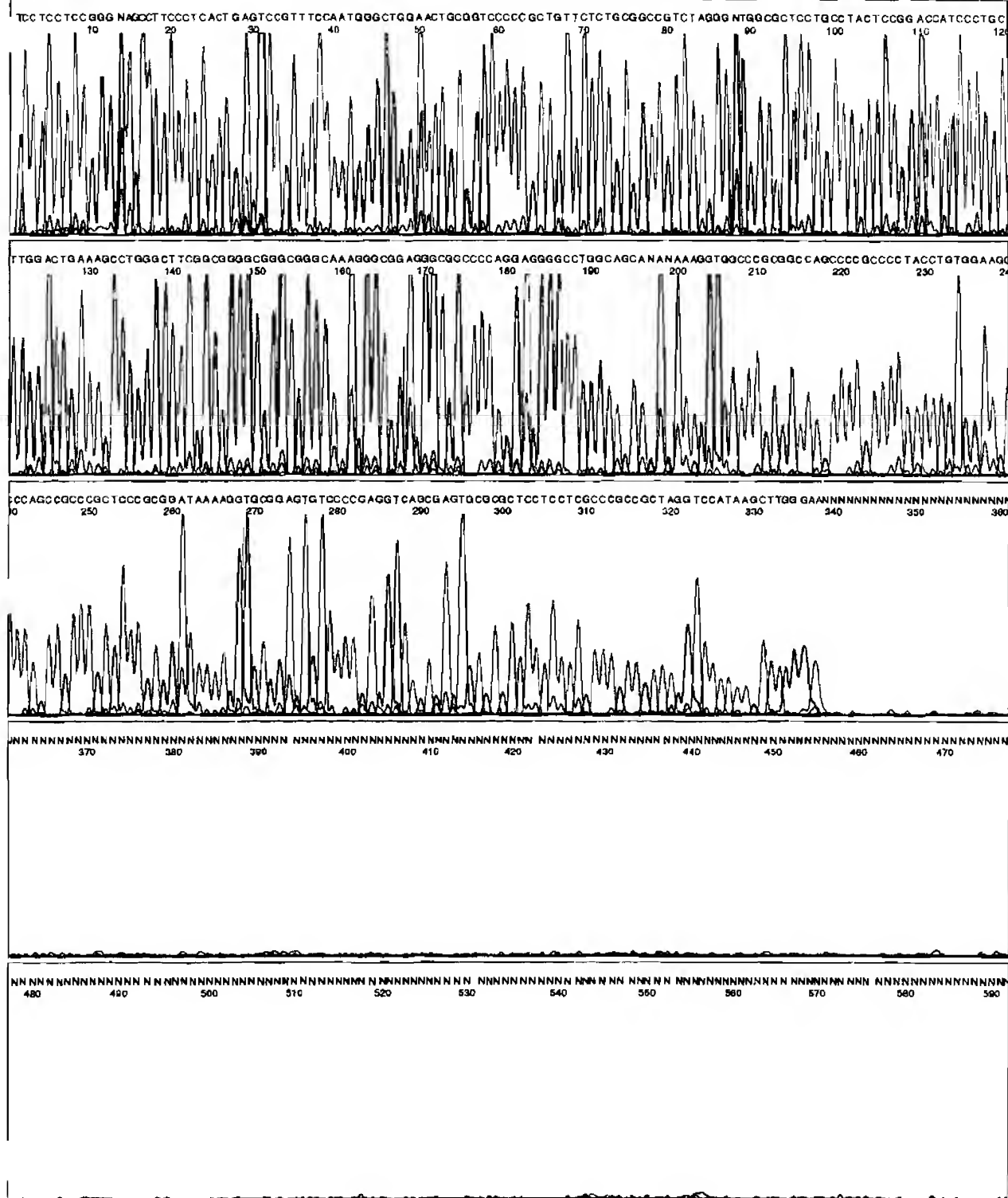
```
Base Call Start: 1323
Base Call End: 11812
Primer Peak Loc.:1323
Signal: G (354), A (393), T (181), C (274)
Matrix Name: Rhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.29 - ABI100
```

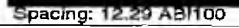
[illegible]

GC383
Lane 19

Rhodamine
Points 1323 to 11812 Base 1: 1323

Spacing: 12.29 ABI100





the 1990s, the number of people in the United States who are 65 years of age or older is projected to increase from 20 million to 35 million (U.S. Census Bureau, 1996).

	1	2	3	4	5	6	7	8	9
1	PCR K7 promoter with Pfu DNA Polymerase								
2									
3	10 μ l 10x buffer								
4	8 μ l 2.5 mM dNTPS								
5	2 μ l template DNA, 100 ng/ μ l								
6	primer		1 μ l	:	pm)	Claw 429			
7			5 μ l	:	"	Claw 430			
8	Pfu. 2 μ l.								
9	H ₂ O 68 μ l.								
10									
11	to two tubes.								
12									
13	94°C. 45"		<div style="display: flex; align-items: center; justify-content: center;"><div style="font-size: 4em; margin-right: 10px;">}</div><div><p>28 Cycles.</p><p>No PCR product!</p></div></div>						
14	94°C. 45"								
15	66°C. 45"								
16	72°C. 1 min								
17									
18									
19									
20	Change annealing Tm:								
21									
22	57°C.		61°C.		30 cycles.				
23									
24	Mix half of the first time volume. - 2 x 25 μ l								
25									
26	Try 57°C and 61°C Annealing. 28 cycles								
27	other condition same. Nothing!								
28									
29									
30									
31									

Insertion of β into pCMV β .

① Cut E/K β from pOX- β with SalI + HindIII.
Isolate ~700bp band, blunt.

② Cut pCMV β with EcoRI + XhoI blunt.

Ligate ① + ②.

Check orientation: cut with HindIII



	1	2	3	4	5	6	7	8	9
1	Do PCR again.								
2									
3	Block 2.				Block 3		Block 1		
4	52°C annealing.				56°C		60°C		
5	# 1. 2. 3.				# 4. 5. 6.		# 7. 8. 9.		
6									
7	1, 4, 7. Tag.				Tag buffer				
8									
9	2, 5, 8. pfu.				pfu buffer.				
10									
11	3, 6, 9. pfu.				Tag buffer.				
12									

PCR Program:

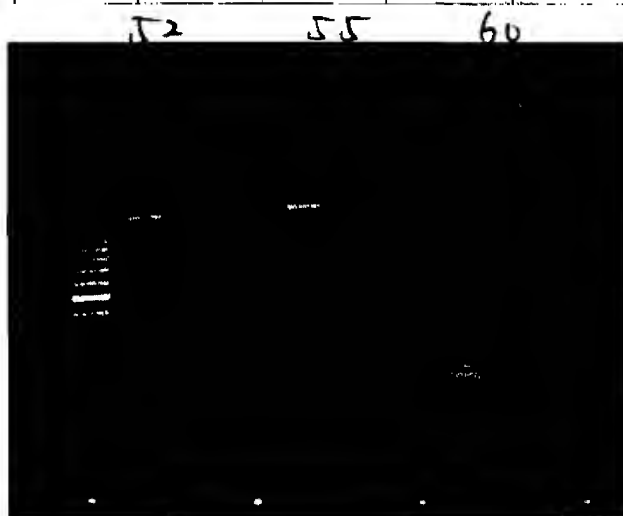
94°C. 2min.

94°C. 45"

Annealing. 45"

72°C. 1'20"

28 Cycles



PCR Mix as before.

↑, ZtoH ↓ (2+5).

+ 41 μl H₂O; 1 μl GC432 (pIND/chop).

+ 2.5 μl Buffer 2; 2.5 μl Buffer 4.

1.5 μl pme. 1.5 μl Hind II. 37°C. 1-15 — 4-15.

Right product should be
~380

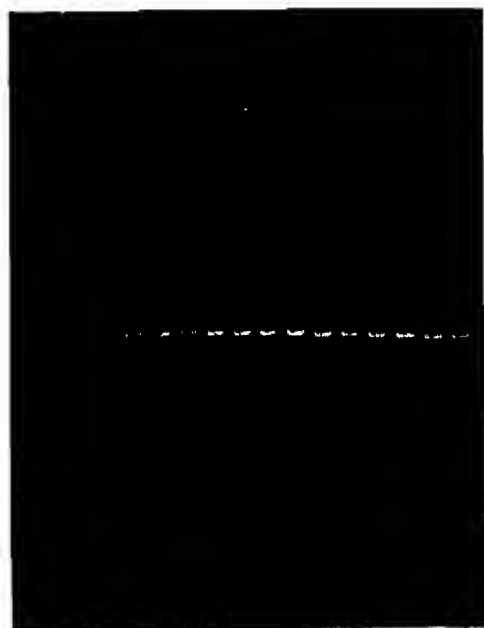


	1	2	3	4	5	6	7	8	9
1									
2		<p> ϕ↑ Z+OH↓. keep in -20°C. </p>							
3									
4									
5									
6		<p> ligation. add 17 μl H₂O. 2 μl ligation Buffer. 1 μl Ligase. RT. 4h. </p>							
7									
8									
9									
10		<p> Transform to DH5α - </p>							
11									
12									
13									
14		<p> Got 10 colonies. PCR screen the </p>							
15									
16									
17									
18		<p> 1-10. primer 387+388. pND/chop control 11-20. " " " </p>							
19									
20									
21									
22		<p> Run on 1.5% agarose gel. </p>							
23									
24									
25									
26									
27									
28									
29									
30									
31									





Redo PCR. Cut together with Gc#32.
 pt. 3 to 4. Ligation. Transform. PCR screen.

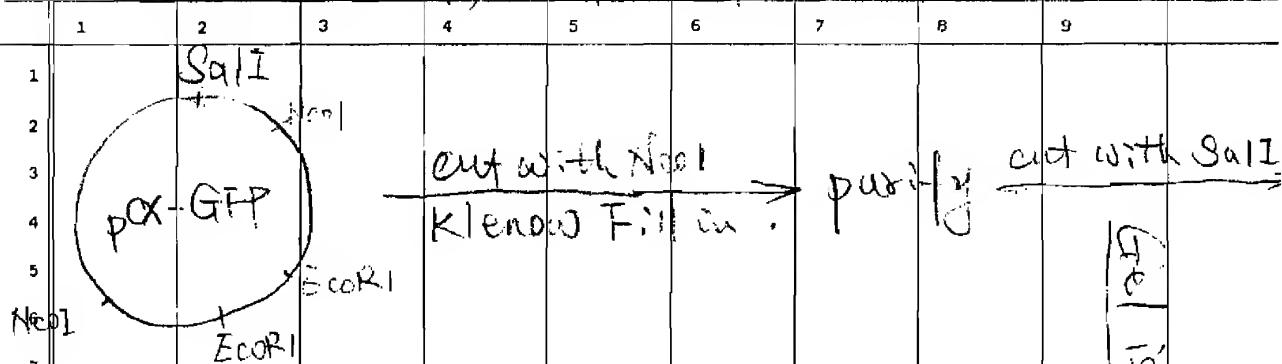


PIND/chop

PIND only

PIND/chop. PCR with Claw 387 + 388.
 product is about 350 bp. After add K7
 promoter, PCR product should be 700⁺ bp.
 L Because there are two pmeI site in
 PIND, plan to clone K7 to pmeI/Hind.
 gap is wrong from beginning. That's why
 B's can't get it. S's design it.

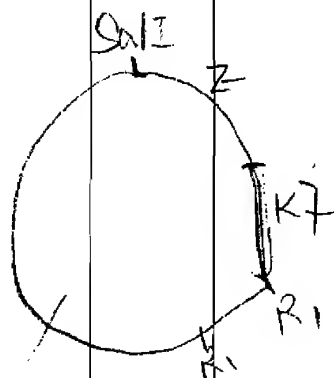
Enhancer / K7 / GFP ... pCX-GFP



From K7/pND. get K7 promoter with pmtI/EcoRI end.

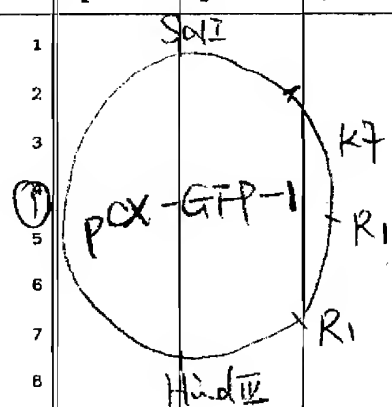
Cut pCX-GFP with SalI, partial cut EcoRI.
keep R1 frag.

Ligation:



pCX-GFP-1

Enhancer / K7 / Snip / pCX-GFP



HindIII SalI + HindIII
gel.

isolate ~700bp band
GeneClean.

② get Snip cassette with HindIII / XbaI ends.

③ cut pCX-GFP, with HindIII. blunt. Ligate.

cut it again with EcoRI. Religate large band.

cut it with SalI + XbaI. isolate large band.

Ligation ① + ② + ③.



Model 377
Version 2.1.1

03-GC387

Signal G:463 A:365 T:294 C:482
DT (BD Set Any-Primer)

Page 1 of 3
9:35 AM

Data Collection

File: 03-GC387
Sample: GC387
Comment:
Lane Number: 3
Channel Number: 31
Number of Scans: 11812
Length: 904
Run started at: 15:44
Run stopped at: 01:44
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1246
Base Call End: 11812
Primer Peak Loc.: 1246
Signal: G (463), A (365), T (294), C (482)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 11.85 - ABI100

```
1 CCCAGCTTTT AGGTGACACT ATAGAATACT CAAGCTATGC ATCAAGCTTT GGAAOCCTGA TGAGTCCGIG 70
71 AGGAAGAAAC GATGACATTC TGCTGACCAG ATTCACGGTC AGCAGAATGT CATGTCGGT TCCAGGATCC 140
141 TTGCTGAAT TCCAAGGGTC TGCGCAACGA CGACGATGAG GTACCAATC GTGTCGTG TGCACTGATG 210
211 AGGCGGTGAG GCGAAACCC TTGACGGGTT CCTATGCGGC CGCTCPAGAG GGCCCAATTC GCGCTATAGT 280
281 GAGTGTATT ACAATTCAT GCGGTGCTT TTACAAAGTC GTGACTGGGA AAACCCCTGGC GTTACCCAAC 350
351 TTAATCGCTT TGCAGCACAT CCCCCCTTCG CCAGCTGGCG TAATAGCGAA GAGGCCCGCA CCGATGCCC 420
421 TTCCAACAG TTGCGCAGCC TGAATGGGA ATGGAAGCGC CCGTAGCGG CGCATTAAGC GCGGCGGGTG 490
491 TGGTGGTTAC GCGCAGGTG ACCGCTACAC TTGCGAGGCG CCGTAGCGCC GCTGCTTTCG CTTTCTTCCC 560
561 TTGCTTTC TCACAGTTCG CCGGCTTTC CCGTCAAGCT CTAAATCGGG GGTCCCTTT AGGGTTCGGA 630
631 TTTAATGCTT TACGGGACCT TCGACCCCA AAAACTTGAT TAGGGTGATG GTTCACGTAA TGGGCCATG 700
701 NCGTATAGA CCGTTTTCG CCGTTTGACG TTGGAAGTC CACGTTCTT AATAAGTGA CTCGTGTTC 770
771 AAATCGAAC AACCATTA CCGTTATCTT GGGGCTATTC NCTTGGATTT TATNANGGA TTTTGCCCGA 840
841 TTTTNGGCC TATTGCGGT AAAAAAATGA ANCTGGNTT TAACCAAAAA TTTTACCGC GNA 910
```

Chop/Perz Right: Save

Class 15



Model 377
Version 2.1.1

26-GC446

Signal G:149 A:117 T:103 C:116
DT {BD Set Any-Primer}

Page 1 of 3
8:46 AM

Data Collection

File: 26-GC446
Sample: GC446
Comment:
Lane Number: 26
Channel Number: 128
Number of Scans: 12992
Length: 1057
Run started at: 16:55
Run stopped at: 03:56
Gel: Gel File
Dyeset/Primer: DT {BD Set Any-Primer}
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1362
Base Call End: 12992
Primer Peak Loc.: 1362
Signal: G (149), A (117), T (103), C (116)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.23 - ABI100

1 GACGCCACT GAATTGTAAT ACGACTCACT ATAGGGCGAA TTGGGCGCTC TAGAGCGGCC GCATAGGAAC 70
71 GCGTCAAGGG TTTCGGGCTC ACGGCCATCAT CAGTGGCGAA CGACGACGAT GTGGTACCTC ATCGTGTGTG 140
141 TTGGCGACAC CCTTGGAAIT CAGGCAAGGA TCCTGGAACC CACGATGACA TTCTGTCTAC CGTGAATCTG 210
211 GTACAGCAGAA TGTTCATCGTT TGTGCTCTAC GGACTCATCA GGGTTCCAAA GCTTGATGCA TAGCTTGAGT 280
281 ATTCTATAGT GTACACCTAAA TAGCTTGGCG TAATCATGGT CATAGCTGTT TCTGTGTGTA AATTGTTATC 350
351 CGCTCACAAT TCCACACAAC ATACGAGCGG GAAGCATAAA GTGTAAAGCC TGGGGTGGCT AATGAGTCTG 420
421 CTAACCTACA TTAATTGGGT TGGGCTCACT GCGCGCTTTC CAGTGGGAA ACCTGTGTGT CCAGCTGCAT 490
491 TATGAATCG GCCAACGGG GGGGAGAGGC GGTTTGGTGA TTGGGCGCTC TTCCGCTTCC TCGCTCACTG 560
561 ACTCGCTGG CTCGGTGTGT CCGCTGGGC GAGCGGTATC AGCTCACTCA AAGCGGTAA TACCGGTAT 630
631 CCACAGATC AGGGGGATAA CGCAAGGAA GAACATGTGA AGCAAAAAGG CCAAGCCAAA AGGCCAGGA 700
701 ACGTAAAAA AGGCCCGGT TGTCTGGGT TTTTCCATAN GNTCCGNC CCCTTGAGC AAGCATTACA 770
-771 AAAAATCGAC GCTTAAAGTC AANAAGGTG GCGAAAAAC CGACAAGGA CTINNTAAAA GATACCCAAG 840
841 GCGTTTTC CCCCCCTGGG AAGGCTTCC CTGTGTGGG CTCTTCTCTT GGTTCGGA ACCCTTGGC 910
911 CGGTTTAAAC CGGGATTACC CTGGTCCGG GNTTNTTTC CCTTTTGGG AAANCCCTTG GNGNCTTTT 980
981 CTINNTATAG NNITAACGN TGTAAAGGNN ATCTTAAAT TTGGGNGTT AAGGTCTGT CNGNTTCCA 1050
1051 AANCTCC 1120

dG - chop / PCR = Right.

Clow 104. Save:

#9



Model 377
Version 2.1.1

19•GC432

Signal G:134 A:105 T:95 C:106
DT (BD Set Any-Primer)

Page 1 of 3
9:02 AM

Data Collection

File: 19•GC432
Sample: GC432
Comment:
Lane Number: 19
Channel Number: 99
Number of Scans: 12992
Length: 1045
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1304
Base Call End: 12992
Primer Peak Loc.: 1304
Signal: G (134), A (105), T (95), C (106)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.06 - ABI100

HindIII

1	GAGAAAGAAG AACTCACACA CAGCTAGCGT TTAAACTTAA GCTTTGGGAC CCTCATGAGT CCGTGAGGAC	70
71	GAAACGATCA CATCTCTGCTG ACCAGATTCA CGGTACAGAG AATGTCATCG TCGGTTCGAG GATCTCTGCC	140
141	TGAATTCCAA GGGTCGTGGC AACGACGACG ATGAGGTACC ACATCGTCTG CGTTGGGCAC TGATGAGGCC	210
211	GTCAGGCCGA AACCCCTGAC GCGTTCCTAT GGGCGGCTC TAGAGGGGCC GTTTAAACCC GCTGATCAGC	280
281	CTCGACTGTG CCTCTCTAGTT GCCAGCCATC TGTGTGTTGC CCTTCCCCCG TGCCCTCCCTT GACCCCTGGAA	350
351	GGTGCCACTC CCACTGTCTT TTCTTAATTA AATGAGGAAA TTGCATGCGA TTGTCTGAGT AGGTGTCATT	420
421	CTATTCTGGG GGGTGGGGTG GGGCAGGACA GCAAGGGGGA GGATTGGGAA GACANNNNNN NGNNTGCTGG	490
491	GGATGNNNGN GGGNNTATG GNTTNTGAGG CNGAAAGAA CCANTGGGNN TTNNGGGGNN NNNNNNNN	560
561	GNNCTGNNNN GGNNTNNNA GNGCGGGNGG NNGNNGGNN NNGNGCAGN GNGCNGNVN NNNNGGNN	630
631	GNNNNNTAG NGNNGNNNN TTNNNNNNN NNNNNNNNC TTNNNGNNN NGNNNGNNN GGNNTNNNN	700
701	GNNNGGNNN NNNNNNNNG NGGNNNNNN TTNNGGGNN NNNNNNNNN NGNNNNNNN NNNNNNNNN	770
771	NNNNNNNNN NNNNNNNNN NGNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN	840
841	NNNNNNNNN TTNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN CNNNNNNNN	910
911	NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN	980
981	NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNNNNNNNN NNCC	1050

pIND / chop . Right !

Save !



Model 377
Version 2.1.1

17-GC430

Signal G:275 A:261 T:228 C:256
DT (BD Set Any-Primer)

Page 1 of 3
9:01 AM

Data Collection

File: 17-GC430
Sample: GC430
Comment:
Lane Number: 17
Channel Number: 90
Number of Scans: 12992
Length: 1057
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1267
Base Call End: 12992
Primer Peak Loc.: 1267
Signal: G (275), A (261), T (228), C (256)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 11.93 - ABI100

```
1  CCCACCTTTT AGGTGACACT ATAGAATACT CAAGCTATGC ATCAAGCTTT GGAACCCCTGA TGAGTCCGIG 70
71  AGGACGAAAC GATGACATTC TGCCTGACCAG ATTCAAGGTC AGCAGAATGT CATGTGTGGT TCCAGGATCC 140
141 TTCAAAGACT AATGAGTCCG TGAGGACGAG ACGAGGATCG AATTCCAAGG GTCTGCCGAA CGACGACGAT 210
211 GAGGTACCAAC ATGTGTGTTCG TTGCGCACTG ATGAGGCGGT GAGGCGGAAA CCTTGACGC GTTCTATGC 280
281 GCGCGCTCTA GAGGGCCCAA TTGCGCTAT AGTGAGTGT ATTACAATTC ACTGGCGGTC GTTTTACAAC 350
351 GTCTGACATG GGAACCCCTT GCGGTACCC AACTTAATCG CCTTGACGA CATCCCTCTT TCGCCAGCTG 420
421 GGTAAATAGC GAAGAGGCC GCACCGATCG CCTTCCCAA CAGTTGGCA GCTGAATGG CGAATGGAG 490
491 CGCCCTGTAG CGGCGCATTA AGCGCGGCG GTGTGGTGT TACGCGCAGC GTGACCGCTA CACTTGCCAG 560
561 CGCCCTAGCG CCGGCTCCTT TCGCTTTCTT CCTTCCTTT CTGCGCACGT TCGCGGCTT TCCCGGTCAA 630
631 AGCTCTAAAT CGGGGCTTC CCTTAGGGT TCGATTTAG TCGCTTACG CACCTCGACC CCAAAAACT 700
701 TGATTAGGT GATGGTTTAC GTAGTGGGCC ATNGGCCCTG ATAGACGGTT TTTCGGCCT TTGACGTGG 770
771 AAGINCACT TTCTTTTAAAT AAGTGGGACT TCTTGGTTTC CAAACTTGG GAACCAACCA CTTTAAACC 840
841 TTATTTTGG GGCCTATTTC TTTTGGATT TAATTAAGG GAATTTTGG CGCAATTTCN GNCCTTNTT 910
911 GGGTINAAAA AAAATGGAGC TTGGATTITA ANCAAAATTT TTAAACCGCG NAAATTTTTA ANCCAAAAT 980
981 TTAAGGGGCG NCCAAGGGG CTTGCTTNA NGGAAAAACC GGAACCCCG TTTGAAAAGG CCCANINCN 1050
1051 CAAAAA 1120
```

pCR II / Chop - pol I - in. Right!



Model 377
Version 2.1.1

07*GC420

Signal G:259 A:240 T:213 C:240
DT (BD Set Any-Primer)

Page 1 of 3
8:59 AM

Data Collection

File: 07*GC420
Sample: GC420
Comment:
Lane Number: 7
Channel Number: 45
Number of Scans: 12992
Length: 1066
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1265
Base Call End: 12992
Primer Peak Loc.: 1265
Signal: G (259), A (240), T (213), C (240)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.01 - ABI100

```
1  CGCCAGCTAT TTAGGTGACA CTATAGATAA CTCAGCTPAT GCATCAAGCT TTGGAACCTT GATGAGTCCG 70
71  TGAGGACGAA ACGATGACAT TCTGCTGACC AGATTACGGT TCAGCAGAAT GTCATCGTUG GTTCCAGGAT 140
141 OCTGCTCTTC TAATGAGTCC GTGAGGAGGA GACGSCCTGA GAATTCCAAG GGTCCTGGCA ACGACGACGA 210
211 TGAGGTACCA CATCGTCGTC GTTGCCTACT GATGAGSCCG TGAGGCCGAA ACCCTTGACG CGTTCCTATG 280
281 CGGCGCTCTT AGAGGGCCCA ATTGCCCCIA TAGTGAGTGG TATTACAATT CACTGGCCGT CGTTTACAA 350
351 CGTGTGACT GGGAAAACCC TGGCGTTAOC CAACHTAATC GCTTGCAGC ACATCCCTT TTGCGCAGCT 420
421 GCGTAAATAG CGAAGAGGCC CGCACCGATC GCGCTTCCCA ACAGTTGGCG AGCTTGAATG GCGAATGGAC 490
491 GCGCCCTGTA GCGGCGCATT AAGCGCGGCG GGTGTGGTGG TTACGCGCAG CGTGACCGCT ACACHTGCCA 560
561 GCGCCCTAGC GCGCGCTCTT TTGCTTTCTT TCCCTTCTTT TCTGCGCAG TTGCGCGGCT TTCCCGTCA 630
631 AGCTCTAAT CGGGGCTCC CTTTAGGCTT CCGATTTAAT GCTTTACGGG ACCTTGACCC CAAAAAACT 700
701 TTGATTAGGG GTGATGGTTC ACGTAAGTGG GNCATTGGCC CTGATAGACG GTTTTTTTCG CCGTTTGACG 770
771 TTGGAAGTCC ACCGTTCTTT TAATAGTGA CTCPTGGTTC CNACTTGGG AACCAACACT TTAAACCTT 840
841 ATTTTNGGC CTATTCTNTT TNGAATTTAT TNANGGAAT TTTTGNCCGA TTTTTCGGG CCGTATNGG 910
911 GTTNAAAAAA ATGGAATTIN GATTTTAAAC CAAAAAANIT TNAACCGCG AATTTTITAA NCCAAAAAT 980
981 TCAAGGGGCG NCCAANGGCG NITGGTTTAA AGGGGAAACC GGGGAAACC CGTTTNAAAA AGGCCAATT 1050
1051 CCCCNAAAA AAACCG 1120
```

PCR II / Chop - B2 - m. Right!



Model 377
Version 2.1.1

09-GC422

Signal G:231 A:216 T:193 C:223
DT (BD Set Any-Primer)

Page 1 of 3
9:00 AM

Data Collection

File: 09-GC422
Sample: GC422
Comment:
Lane Number: 9
Channel Number: 53
Number of Scans: 12992
Length: 1069
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1265
Base Call End: 12992
Primer Peak Loc.: 1265
Signal: G (231), A (216), T (193), C (223)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.01 - ABI100

```
1  CGCCAGCTAT TTAGGTGACA CTATAGAATA CTCAGCTAT GCATCAAGCT TIGGAACCCG GATGAGTCCG 70
71  TGAGGACGAA ACGATGACAT TCTGCTGACC AGATTGACGG TCAGCAGAAT GTTCATGTCG GTTCCAGGAT 140
141 CCTCGAAGCT GTCTGATGAG TCCGCTGAGG CGAAACCGCG TTGAGAAATC CAAGGGTCTG CGCAACGACG 210
211 ACGATGAGGT ACCACATGCT CGTCTGTGCG CACTGATCAG GCGGTGAGGC CGAAACCCCT GACGCGTTC 280
281 TATGCGGCGC CTCTAGAGGG CCCTAATGCG CCTATAGTGA GTCTGATTAAC AATTCACCTG CCGTCTGTTT 350
351 ACAACGTCGT GACTGGGAAA ACCCTGGCGT TACCCAACTT AATCGCCCTG CAGCACATCC CCTTTTGGCC 420
421 AGCTGGCGTA ATAGCGAAGA GCGCGGCACC GATCGCCCTT CCCAACAGTT GCGCAGCCCT AATGGCGAAT 490
491 GGACGCGGCC GTGTAGCGCG CATTAAGCGC GCGGGGTTG GTGGTTACGC GCAGCGTGAC CGCTACACTT 560
561 GCGAGCGGCC TAGCGCGCGC TCCCTTCTGT TTCTTCCCTT CCTTTCTGCG CAGGTTCGCG GCGTTTCCCG 630
631 GTCAAAGCTC TAAATCGGGG GCTCCCTTTA NGGTTCGGAT TTAATGCTTT ACGGNACTT GACCCCAAAA 700
701 AACTTGATTA GGGTGATGGG TTACGTTTAG TGGGCCATCG CCTGATAGA CGGTTTTCG GCGTTTTCGAC 770
771 GTTTGGAAGT CCACGTTCTT TTAATAGTGT GACTCTTGGT TCCAAACCTT GGGAAACCAAC CACTTTTAAAC 840
841 CCTTATTTT TNGGGCTAAT TCCCTTTTGG AATTTAATTA NGGGAATTTT TGGCGGATTT TNGGGGCTTA 910
911 TTTGGGGTTA AAAAAAATGG AAGCTTGGAT TTTTAANCCA AAAAAATTTT TAACGCGCGA AATTTTAAAC 980
981 CCAAAAAATT TCAAGGGGCG CCNAAAGCGC NTGTNTTAA AGGGNAAAC CGGGAACCCG CTTTNNAAAA 1050
1051 AGGCCAGTT NCCCNANT
```

pCR1/chop - C3

Right:



Model 377
Version 2.1.1

10•GC423

Signal G:219 A:202 T:185 C:202
DT (BD Set Any-Primer)

Page 1 of 3
9:00 AM

Data Collection

File: 10•GC423
Sample: GC423
Comment:
Lane Number: 10
Channel Number: 58
Number of Scans: 12992
Length: 1058
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1292
Base Call End: 12992
Primer Peak Loc.: 1292
Signal: G (219), A (202), T (185), C (202)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.02 - ABI100

```
1  CCAGCTATTT AGGTGACACT ATAGAATACT CAAGCTATCC ATCAAGCTTT GGAACCCCTGA TGAGTCCGTG 70
71  AGGACGAAAC GATGACATTC TGCTGACCAG ATTACACGGTC AGCAGAATGT CATCGTCGGT TCCAGGATCC 140
141  TCGAAGCTGT CTGATGAGTC CGTGAGGACG AAACCGGGTT GACAATTCCA AGGGTCTGGG CAACGACGAC 210
211  GATGAGGTAC CACATCGTTC TCGHTGCGCA CTGATGAGGC CGTGAGGCGG AAACCCCTGA CGCGTTCCTA 280
281  TCGGGCGGCT CTAGAGGGCC CAATTGCGCC TATAGTGAGT CGTATTACAA TTCACTGGCC GTCGTTTTC 350
351  AACGTCTGA CTGGGAAAAC CCTGGCGTTA CCCAACTTAA TCGCCTTGCA GCACATCCCC CTTTCGCCAG 420
421  CTGGCGTAAT AGCGAAGAGG CCGGCACCGA TCGCCCTTCC CAACAGTTGC GCAGCCTGAA TCGCGAATGG 490
491  ACGCGCCCTG TAGCGCGCA TTAAGCGCGG CCGGTGTGGT GGTACCGGC AGCGTGAACG CTACACTTGC 560
561  CAGCGCCCTA GCGCCCGCTC CTTTCGCTTT CTTCCCTTCC TTCTCTGCA CGTTGCGCG CTTTCGCCGT 630
631  CAAGCTCTAA ATCGGGGGCT TCGTTTAGG TTCCGATTTA GTGCTTTACG GNACTTGAC CCCAAAAAC 700
701  TTGATTAGGG TGATGGTTC CGTAGTGGGC CATTGGGCTG ATAGAACGGT TTTTGGGCC TTTTGACGTT 770
771  TGGAGTTCCA ACGTCTTTT AATAGTGGAC TTCTTTGGTT CCCAACTGG GAACCAACCA CTTTAAACCC 840
841  TTATTTTGG GNCATTTTC CTTTTCGAA TTTAATTAG GGAATTTTG GCGCGATTT TCGGGGCTT 910
911  TTTCGGGTIN AAAAAAATG GAGCTTGANT TTTAACCAAA AATTTTINAA CCGCGCAAN TTTTAAACCA 980
981  AAAATTTINA GGGGCCCCA ANGGGCTTG NTTTAAAGG GAAACCGGG AACCCCNTE TAAAAAGCCC 1050
1051 AATTCCCC 1120
```

pCRI/chop - 43 Right!



Model 377
Version 2.1.1

11•GC424

Signal G:245 A:228 T:202 C:231
DT (BD Set Any-Primer)

Page 1 of 3
9:00 AM

Data Collection

File: 11•GC424
Sample: GC424
Comment:
Lane Number: 11
Channel Number: 62
Number of Scans: 12992
Length: 1057
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1280
Base Call End: 12992
Primer Peak Loc.: 1280
Signal: G (245), A (228), T (202), C (231)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.01 - ABI100

```
1  NCCAGCTATT TAGGTGACAC TATAGAATAC TCAAGCTATG CATCAAGCTT TGGAAACCCIG ATGAGTCCGT 70
71  GAGGACGAAA CGATGACATT CTGCTGACCA GATTCACGGT CAGCAGAATG TCATOGTCCG TTCCAGGATC 140
141 CTCTTCGACT GATGAGTCCG TGAGGACGAA ACATGGCTGA ATTCCAAGGG TCTGCGCAAC GACGACGATG 210
211 AGGTACCACA TCGTCGTCTG TCGCACTGA TGAGSCCGTG AGGCGGAAAC CCTTGACGCG TTCCATGCG 280
281 GCGCTCTAG AGGCGCCAAT TCGCCCTATA GTGAGTGTGA TTACAATTCA CTGGCGCTCG TTTTACAACG 350
351 TGTGACTGG GAAAACCCIG GGGTTACCA ACITTAATGC CTTCGAGCAC ATCCCCCTTT CGCCAGCTGG 420
421 CGTAATAGCG AAGAGGCGCG CACCGATCGC CCTTCCCAAC AGTTGCGCAG CCTGAATGCC GAATGGACGC 490
491 GCGCTGTAGC GCGCGATTAA GCGCGGCGGG TGIIGTGGTT ACGCGCAGCG TGACCGGTAC ACTTCCAGC 560
561 GCGCTAGCGC CCGCTCCCTT CGCTTCTTTC CCTTCCCTTC TCGCACGTTT CGCGGCTTTT CCGCTCAAG 630
631 CTCTAAATCG GCGCTCCCTT TTAGGGTTCC GATTTAGTGC TTTACGGGAC CTNGACCCA AAAAAGTTGA 700
701 TTAGGGTGAA TGGTTACAGT AGTGGGCGCA TTGCGCTTGA TANACGGTTT TTTGCGCCTT TGGACCTTIG 770
771 GAAGTCCACG TTTTCTAAT AGNGGACNTT TTGGTTCCAA AACTGGNACC AACNANTINA ACCCTATTTT 840
841 TGGGTCTAAT TCTTTTGGCA ATTTATTAAG GGGATTTTIG GCGCGATTTT CCGGCGCTNT TGGGGTINAA 910
911 AAAAAATGGA ACCTTGATTT TNACCCAAAA AATTTTAAAN CCGCGAAATT TTTTAACCA AAAATTTTANG 980
981 GGGCCCCAAA GGGGCTTTTG NTTTAAGGG GAAACCGGG AACCCCTTTN AAAAAAGGCC TTTCGCCNAA 1050
1051 AAAANCG
```

PCR II / chop - C9 Right!



Model 377
Version 2.1.1

13-GC426

Signal G:288 A:270 T:244 C:265
DT (BD Set Any-Primer)

Page 1 of 3
9:00 AM

Data Collection

File: 13-GC426
Sample: GC426
Comment:
Lane Number: 13
Channel Number: 72
Number of Scans: 12992
Length: 1041
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1280
Base Call End: 12992
Primer Peak Loc.: 1280
Signal: G (288), A (270), T (244), C (265)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.02 - ABI100

1	CCAGCCTATT	AGGTGACACT	ATAGAACTACT	CAAGCTATGC	ATCAAGCTTT	GGAAACCCGA	TGAGTCCGTG	70
71	AGGACGAAAC	GATGACATTC	TGCTGACCAG	ATTCAACGGTC	AGCAGAATGT	CATCGTCCGT	TCCAGGATCC	140
141	TCTTCGACTA	ATGAGTCCGT	GAGGACGAGA	CATGGCTGAA	TTCCAAGGGT	CTGCCCAACG	ACGACGATGA	210
211	GGTACCATAT	CGTCGTGCTT	GCGCACTGAT	GAGGCGGTGA	GGCCGAAACC	CTTGACCGGT	TCCATATGCG	280
281	CCGCTCTAGA	GGGCCCAATT	CGCCCTATAG	TGAGTCTGAT	TACAATTCAC	TGGCCGTCTT	TTTACAACTG	350
351	CGTGACTGGG	AAAACCCCTG	CGTTACCCAA	CCTAATCGCC	TTGCAGCACA	TCCCCCTTTC	GCAGCTGGC	420
421	GTAATAGCGA	AGAGGCCCCG	ACCGATCGCC	CCTCCCAACA	GTTCCGCAGC	CTGAATGGCG	AATGGACCGG	490
491	CCCTGTAGCG	GCGCATTAAG	CGCGGCGGGT	GTGGTGGTTA	CGCGCAGCGT	GACCGCTACA	CTTGCCAGCG	560
561	CCCTAGCGCC	CGCTCCTTTC	GCTTCTCTCC	CCTCCCTTCT	CGCCAGGTTT	GCCGGCTTTC	CCCGTCAAGC	630
631	TCTAAATCGG	GGCTCCTCTT	TAGGGTTCGG	ATTTAGTCTG	TTACGGNACC	TCCACCCCAA	AAAACCTGAT	700
701	TAGGGTGATG	GGTCAAGTAG	TGGGCCATCG	CCCTTGATAG	ACGGTTTTTC	GGCTTTTGAC	GTTCGAAGIN	770
771	CACGTTCTTT	AATAGTGGAC	TTCTTGCTTC	CAAACITGGG	ACAACACTTN	AACCCCTATC	TTTGGNCIAT	840
841	TCTTTTIGAA	TTTATTTAAG	GGAATTTTGC	CCGATTTTCC	GGGCTATTIN	GGGTINAAAA	AAATGGAAGC	910
911	TTGANTTTTA	ACCAAAAAAT	TTTAAACCGC	GGAAATTTTT	AACCAAAAAAN	TTTCAAGGGG	CCCAAGGGG	980
981	CTTCCTTTAA	AGGGGAANCC	GGGAACNNCC	TTTINAAAAAG	GCCAGINCC	GCAANAAAAAN	G	1050

PCR II / chop - C9 - m Right!



Model 377
Version 2.1.1

15•GC428

Signal G:316 A:288 T:245 C:276
DT (BD Set Any-Primer)

Page 1 of 3
9:01 AM

Data Collection

File: 15•GC428
Sample: GC428
Comment:
Lane Number: 15
Channel Number: 81
Number of Scans: 12992
Length: 1079
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1268
Base Call End: 12992
Primer Peak Loc.: 1268
Signal: G (316), A (288), T (245), C (276)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.01 - ABI100

```
1  CACCAGCTAT TTAGGTGACA CTATAGAATA CTCAGCTAT GCATCAAGCT TTGGAACCTT GATGAGTCCG 70
71  TGAGGACGAA ACGATGACAT TCTGCTGACC AGATTCAOGG TCAGCAGAAT GTCACTGTCG GTTCCAGGAT 140
141 CTTTCAAAGA CTGATGAGTC CGTGAGGACG AAAACGAGGAT CGAATTCCAA GGGTCTGCGC AACGACGACG 210
211 ATGAGGTACC ACATGCTCGT CGTTGCGCAC TGATGAGGCC GTGAGGCCGA AACCTTTCAC GGGTTCCTAT 280
281 GCGGCGCTC TAGAGGCGCC AATTGCGCCT ATAGTGAGTC GTATTACAAT TCACTGGCGG TCGTTTACAA 350
351 ACGTGTGAC TGGGAAAACC CTGGCGTTAC CCAACTTAAT CGCTTTCAG CACATCCCCC TTTCGCCAGC 420
421 TGGGTAAATA GCGAAGAGGC CGGCACCGAT CGGCTTCCG AACAGTTGCG CAGCTTGAAT GCGAATGGA 490
491 CGCGGCTGT AGCGGCGCAT TAAGCGCGGC GGGTGTGGTG GTTACGCGCA GCGTGACCGC TACACTTGCC 560
561 AGCGGCTAG CGCGGCTCC TTTCGCTTTC TTCCCTTCTT TTCTCGCAC GTTCGCGGCG TTTCGCGGTC 630
631 AAGCTCTAAA TCGGGGGCTC CCTTTAGGGT TCCGATTAA GTGCTTTACG GNACCTGAC CCCAAAAAAC 700
701 TTGATTAGGG TGATGGGTTC ACGTAAATGG CATCGGCTT GATGACGGG TTTTTCGCGC TTTTTCAGTT 770
771 GGAAGTTCCA CGTTCTTTT AATAGTGGGA CTCTTTGGTT CCAAACTTG GGAACCAAC ACTTTAAACC 840
841 CCTTATTTT NCGGCTAAT TCCTTTNGA APTTAATTAA GGGGAATTT TGGCGAANT TTGNGGCT 910
911 TATTTGGGT TAAAAAAA TGGAANCITG GATTTTNAAC CCAAAAAAN TTTTAAACG CGGAAATTT 980
981 TTTAANCCAA AAATTTCAAN GGGGCGNCCA ANGGGCGCTT GGTTTTNAAG GGGAAACNG GNAACCCCT 1050
1051 TNNAAAAAG NCCAATTNCC CCAAAAAA 1120
```

PCR II / Chop - Pol II Right!

Seize!



Model 377
Version 2.1.1

05-GC418

Signal G:263 A:241 T:214 C:236
DT (BD Set Any-Primer)

Page 1 of 3
8:59 AM

Data Collection

File: 05-GC418
Sample: GC418
Comment:
Lane Number: 5
Channel Number: 36
Number of Scans: 12992
Length: 1091
Run started at: 15:25
Run stopped at: 02:26
Gel: Gel File
Dyeset/Primer: DT (BD Set Any-Primer)
Comb: 36-well sharks-tooth
Instrument Name: 377
Collect Vers.: N/A

Data Analysis

Base Call Start: 1265
Base Call End: 12992
Primer Peak Loc.: 1265
Signal: G (263), A (241), T (214), C (236)
Matrix Name: dRhodamine
Channels Ave.: 3
Analysis Vers.: Version 2.1.1
Base Spacing: 12.00 - ABI100

PCR1

HindIII

BamHI

XbaI

1	CNCCAGCTAT	TTAGGTGACA	CTATAGAATA	CTCAAGCTAT	GCATCAAGCT	TTGGAACCT	GATGAGTCCG	70
71	TGAGGACGAA	ACGATGACAT	TCTGCTGACC	AGATTCAAGG	TCAGCAGAAT	GTCATGCTCG	GTTCCAGGAT	140
141	CTTGCTCTTC	TGATGAGTCC	GTCAGGACGA	AACCGCTCGA	CAATTCCAG	GGTCTGCGCA	ACGACGACGA	210
211	TGAGGTACCA	CATGCTGCTC	GTTGCGCACT	GATGAGGCGG	TGAGGCGGAA	ACCCCTGACG	CGTTCTATG	280
281	CGGCGCTCT	AGAGGGCCCA	ATTGCGCCTA	TAGTGAGTGG	TATTTACAAT	CACCTGCGCT	CGTTTTACAA	350
351	CGTGTGACT	GGGAAAACCC	TGGCGTTACC	CAACTTAATC	GCCCTGCAGC	ACATCCCCCT	TTCGCCAGCT	420
421	GGCGTAATAG	CGAAGAGGCC	CGCACCGATC	GCCCTTCCCA	ACAGTTGCGC	AGCCCTGAATG	GCGAATGGAC	490
491	GCGCCCTGTA	GCGGGCCATT	AAGCGCGGCG	GGTGTGGTGG	TTACGCGCAG	CGTGACCGCT	ACACTTGCCA	560
561	GCGCCCTAGC	GCCCGCTCTT	TTCGCTTTCT	TCCCTTCTT	TCTGCGCAG	TTCGCGGCT	TTCGCGGCT	630
631	AGCTCTAAAT	CGGGGCTTCC	TTTAGGGTTC	CCGATTTAAT	GCTTTACGGG	ACCTTGACCC	CAAAAACTT	700
701	GATTAGGGTG	ATGGTTCACG	TAATGGGGCC	ATNGNCTTG	ATAGACGGTT	TTTTGNCTT	TTGACGTTG	770
771	GNAGINCAEC	GTTCNTTTIN	AATAGTGGGA	CCNTTTGGIT	TCCCAAACCT	GGGAACCAAA	CAACTTTAAA	840
841	NCCCTTTTTC	TTTGGGGCCT	AATTCCTTTT	TGGANITTTA	TINAAGGGGG	ATTTTGTGGC	CCGAATTTTC	910
911	NGGGCTTTT	TTGGGGTTAA	AAAAAAATGG	GAGCTTGGGA	TTTTTAACCA	AAAAANITTT	TAAACCGCGG	980
981	AAATTTTTTA	ACCAAAAAAT	TTCANGGGGC	NCCCAAAGGG	GGCTTTGNTT	TAAAGGGGAA	AACCGGGAAA	1050
1051	CCCCCTTTTA	AAAAGGGGCC	ATTTCCCCN	AAAAAAACCN	G			1120

PCR1/chop-B2

right!

Chop #3: HindII - BglII - StyI - XbaI.

Ge isolate PCR #1 + (Chop)

PCR #2 + #4 (dG - chop)

primers

templet

1. 413 + 414

> pNewclip

57°C Annealing.

2. 413 + 415

3. 413 + 414

> pChop #3

94°C 30"

4. 413 + 415

57°C 40"

5. 431 + 433

> pNewclip

72°C 1'

6. 432 + 433

7. 431 + 433

> pChop #3

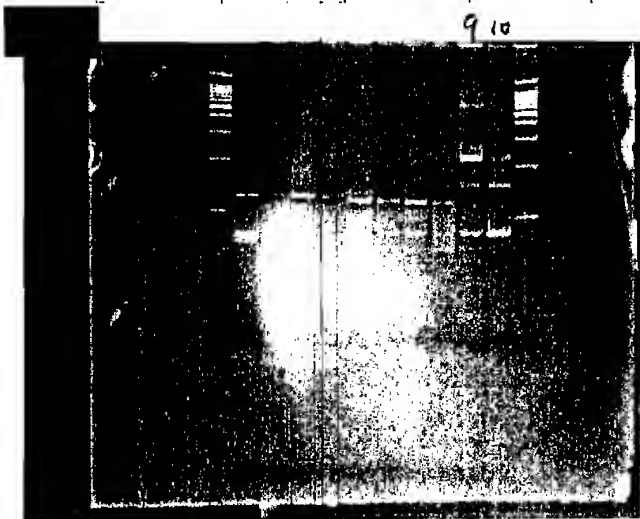
8. 432 + 433

9. 413 + 433

Chop temp

10. 413 + 433

dG chop temp.



gel purity fusion bands 9 and 10



PCR screen condition:

10 p m 104. 1 M
 " 105 1 M.
 1 M 4 mM dNTP.
 Tag 14/x 1 M
 Buffer 2 M
 MgCl₂ 25 mM. 2 M.
 DNA. 1 M (cell 8)
 H₂O 1 M

94°C 30"

52°C 40"

72°C 60"

28 cycle.

Run 1.6% gel check:

Marke. 9x1746 M. 1-14. per PCR.
 " " 15-28 "

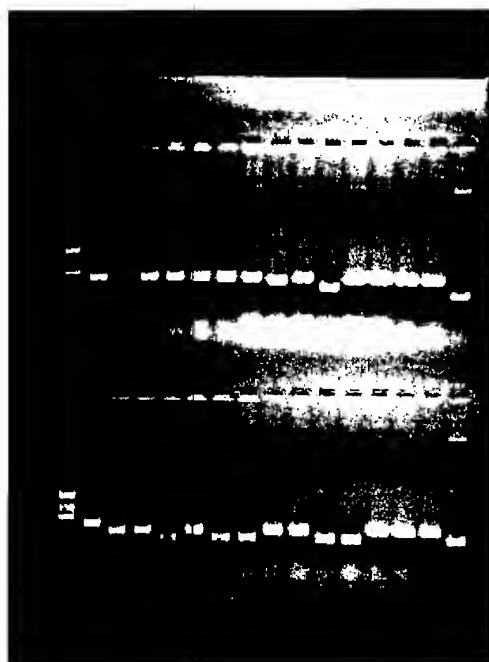


IMAGE SIZE (640 x 400 x 8).
 INT PERIOD = 0.46 SEC.
 ACQUIRED

STRAT-GENE EAGLE EYE II
 21:56:44

Chop:

1. 3. 4. 5. 6.

dg - Chop.

7. 9. 11. 12.

13. 14. 15.

maybe right.

Zuo en late to
 1 R + A. D

Isolate plasmid. Cut them with
 BamH₁ or EcoR₁ old Chop #3 as
 Control. (cut 3.5M)

Lane order:

BamH₁ 1, 3, 4, 5, 6, 7, 9, 11, 12, 13, 14, 15, old #3, Marker

EcoR₁

#3. old

8u1
 Hind@Eco

BamH₁

EcoR₁

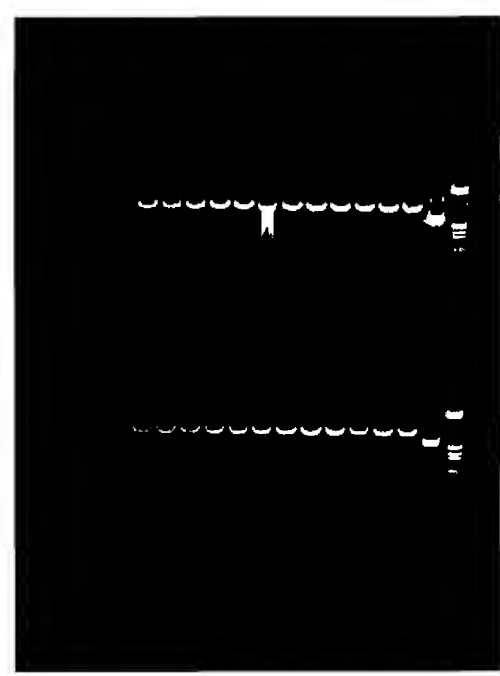
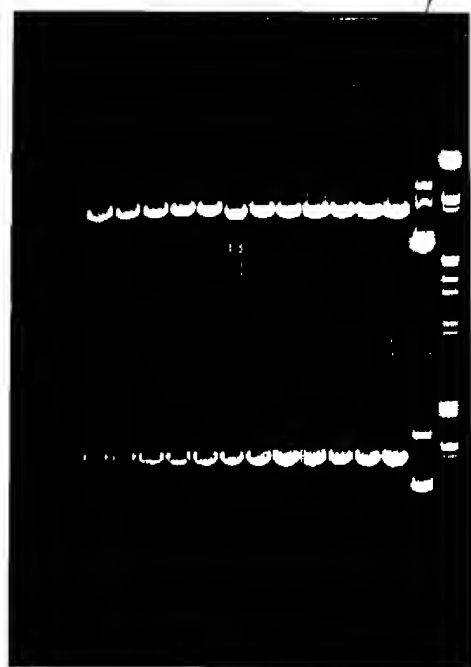


IMAGE SIZE (mm) 480 x 87.
 INT. PERIOD 0.17 SEC.
 -COPIED
 STRATGE EOLE EYE II 28:15:15

old #3 is : HindII - BglII - SstI - XbaI. Can't be a
 all of the candidate can be cut. Sent to

Put C3, C9 B2 pol2 to chop / PCR

Chop #4. GC387 is right! Finally!
Inoculate it to 25 ml LB + Amp

Make stocks: PCR / Chop. 1/98.

Isolate plasmid. 4 miniprep. Final
320 M. 0.2 μ g/ λ .

Cut with BamH₁ + EcoR₁:

1. 30 μ l DNA + 30 μ l H₂O + 7 μ l NEB2 1.5 μ l BamH₁
1.5 μ l EcoR₁

* 2. 20 μ l sequencing left.

1% SeaPlaque gel isolate GeneClean,
Final inoc H₂O

Use 2 Ligation

				Buffer	Ligase
1	B2	1-10.5 μ l	+ 2 μ l V	1.2 μ l	0.6 μ l
2	B2-m	"	"	"	"
3	C3	"	"	"	"
4	C3-m	"	"	"	"
5	C9	"	"	"	"
6	C9-m	"	"	"	"
7	pol1	"	"	"	"
8	pol1-m	"	"	"	"
9	chop/PCR	HindIII/XbaI Vector only	#2. 2 μ l	"	"
	add H ₂ O to	12 μ l	18°C. 0/h		



	1	2	3	4	5	6	7	8	9
1									
2									
3									
4									
5									
6									
7									
8									
9									
10									
11									
12									
13									
14									
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22									
23									
24									
25									
26									
27									
28									
29									
30									
31									

transform 5/7 ligation stuff to DH5X.

PCR Screen

B2 : 1-6

B2-m : 7-12

C3 : 13-18

C3-m : 19-24X

C9 : 25-28

C9-m : 31-36

pol2 : 37-39

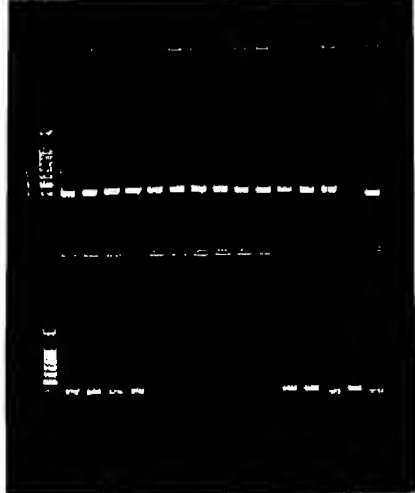
pol2-m : 40-42

29, 30. PERE/chop V.

387.388. PIND/chop : 47-56, 57 = PINDV.

Run gel.

M. 1 - 14 29



#14 is wrong.

#19-24, all of the C3-m are wrong.

#27 is wrong.



	1	2	3	4	5	6	7	8	9
1	Screen the left.							PCR II/chop.	
2									
3	100bpM.	31-36.	V.	37-42.	V.				
4									
5	M. 100bp	47-57.		100bpM.					
6									

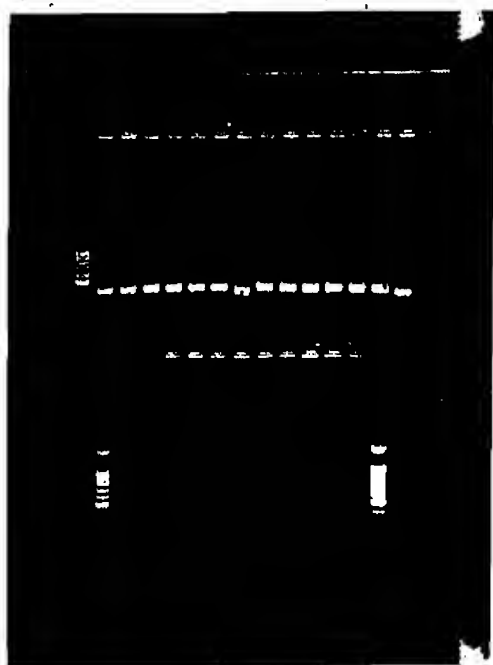


IMAGE SIZE (640 x 480 x 81)
INIT PERIOD = 5.49 SEC.
ACQUIRED

SYNTHASE END EYE II 17:27:28

22
23
24
25
26
27
28
29
30
31

Inoculate these to 4ml LB + Amp:

B2 = 1, 2.

B2-m = 7, 8.

C3: 13, 15.

C9: 25, 26.

C9-m: 31, 32.

pol2: 37, 38.

pol2-m: 40, 41.

Isolate plasmids

OD₂₆₀:

according to Seq. res
Same

B2	#1	0.08232	GC 418
	2	0.09574	419
B2-m	7	0.08361	420
	8	0.09139	421
C3	13	0.11688	422
	15	0.11341	423
C9	25	0.09106	424
	26	0.11543	425
C9-m	31	0.09956	426
	32	0.10767	427
P012	37	0.04975	428
P012	38	0.11219	429
P012-m	40	0.08922	430
	41	0.12112	GC 431

Sent to be sequenced using Clon105

PIND/Chop.	#50	0.09663	GC 432	✓
PIND/Chop	#51	0.10231	GC 433	✓